CAN OPSONINS BE OBTAINED DIRECTLY FROM BACTERIA AND YEASTS?

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The curve of the opsonic indices of an individual who has been treated with a bacterial vaccine, shows soon after inoculation a fall and a subsequent rise, after which the curve remains at a level higher than it had before the inoculation. The falling and rising have been called by Wright the negative and positive phases. It is reasonable to suppose that the injected microbe gives out a substance, either an anti-opsonin or an anti-phagin, which is responsible for the negative phase. We know that saline does extract such a substance from bacteria.* But it is not known if, subsequent to the liberation of all the anti-phagin, the bacteria give off the opsonin which produces the positive phase. From what we believe to be the mechanism of immunisation, it is probable that the opsonin is not derived directly from the bacteria but rather from the body-cells in response to the anti-phagin. On the other hand we know that the ingestion of yeast, from which I have not been able to obtain evidence of the secretion of an anti-opsonin, leads to the production of a certain amount of immunity against staphylococcus. The digestion of the yeast would therefore appear to give rise to opsonin, and such being the case, it is difficult to think otherwise than that the digestion of the bacteria would bring about the same result. We know that bacteria and yeast are comparatively rich in nucleoproteid, and we have it from Busse that nucleic acid protects the individual against the invasion of staphylococci and *B. coli*.

*These Proceedings, 1908, p.669. As the anti-body is not destroyed at 70° it is not aggressin.

In an attempt to detect the liberation of opsonin from bacteria under the influence of pepsin-hydrochloric acid, ferric chloride was added to assist digestion. Experiments had shown that a greater loss of staining power as indicated by the Gram method of staining was obtained when to the pepsin-hydrochloric acid, salts of iron, calcium, barium, nickel or aluminium had been added, and of these salts ferric chloride appeared to be most active.

The results of a great number of experiments made with yeast and with *Staphylococcus aureus* which in some cases had been heated in saline at 60° for an hour, showed that no opsonin was liberated. The bacteria, either when heated or not, continued to give off anti-opsonic bodies for a considerable time.

In experiments such as these it is necessary to have the digested extracts absolutely free from bacterial cells. This condition was not always obtained upon neutralising the acid ferric chloride with sodium carbonate as the precipitate of ferric hydrate appeared to be too rapidly formed to entangle all the bacterial cells, and an apparently brilliant fluid contained enough cells to vitiate the results. A clear and trustworthy fluid was obtained by treating the first clear extract, freed from iron, with calcium chloride and phosphoric acid and neutralising with sodium carbonate. The precipitate of calcium phosphate is formed slowly and entangles all the partly digested bacteria.

Subsequent treatment of the bacteria with pancreatic extract (liquid pancreatin from Parke, Davis & Co.) in faintly alkaline solution or in 0·2% Na_2CO_3 also gave negative results.

Yeast-extract as obtained by grinding up yeast with sand and extracting the mass with a small quantity of saline also showed no trace of opsonin.

It is concluded from the research that opsonins are not directly obtainable from either bacteria or yeast.